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PERITONITIS CAUSED BY THE PROTEUS VULGARIS.

By Simon Flexner, M. D., *Associate in Pathology.*

(*From the Pathological Laboratory of the Johns Hopkins University and Hospital.*)

presented by the author

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Hauser, in 1885, isolated from putrefying animal substances the proteus group of micro-organisms, and these possessed the property of setting up putrefactive decomposition when added in pure culture to sterilized or non-sterilized meat. A number of instances have been published since which tend to show that the proteus bacteria may exert pathogenic effects on human beings.

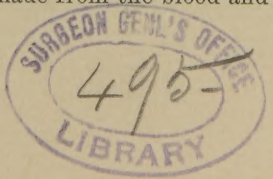
Foà and Bonome found in the blood and organs of a man dead of supposed volvulus with hemorrhagic infarction of the intestine and mesentery and thrombosis of the mesenteric vein, a bacillus which they identified as the proteus vulgaris.

Hlava found the same organism in a case of "hemorrhagic infection," but he regarded it as accidental and ascribed no pathogenic effects to it.

Hauser described a case of phlegmonous inflammation of the hand and forearm from which he obtained both proteus and streptococcus. He considers that an actual symbiosis exists between the two bacteria, and thinks the necrosis of tissue caused by the streptococcus enabled the proteus to grow, while the effect of the latter in diminishing the tissue resistance permitted the streptococcus to invade more readily.

Proteus has been found in the intestinal canal of infants suffering from summer diarrhœa, by Booker and by Baginsky, and it is regarded by them as being of pathological significance.

The recent work of Jaeger, if confirmed, proves that the proteus organisms possess marked pathogenic properties. He studied a series of cases of Weil's disease which occurred in the garrison at Ulm. His investigation comprised eight cases, two of which came to autopsy. The remaining six were studied during life, cultures being made from the blood and urine.



In the two cases which were examined at autopsy no organisms were found in cover-slip preparations from the organs. But agar-agar, gelatine and bouillon cultures from the liver, kidneys, spleen and medulla oblongata of the first one, and liver, spleen and kidneys from the second, gave positive results. The organisms cultivated from these sources were identified with the proteus group. Examination of sections, stained in Kühne's methylene-blue, gave positive results. As a rule the organisms were more difficult to demonstrate in the kidneys than in the other organs, but when found they were often present in surprising numbers.

The blood of the six cases gave negative results, while the urine, drawn into sterilized Erlenmeyer's flasks after thorough cleansing and disinfection of the penis, gave in cover-slip preparations a positive result in every case. Cultures made from the urine were successful in four of the six cases. The organisms obtained were identified with proteus.

Babes reports the case of a boy 13 years old who exhibited uræmic symptoms and diarrhœa, and at the autopsy was found to have cirrhosis of the liver, hydronephrosis and extensive diphtheritic necrosis of the mucous membrane of the large intestine. From the organs of this case he obtained a bacillus which he regarded as proteus vulgaris.

The case of proteus infection which we observed was that of a young woman aged 18, white, who was admitted into the service of Dr. Osler, Johns Hopkins Hospital, on October 29th, 1892, and died on October 31st. When admitted, she was irrational and answered questions in a confused way. She complained of pain in the abdomen. No history could be obtained.

Status præsens. She has some twitching of the extremities. She slept well during the first part of the night, but after that was restless; this morning she vomited. Face rather flushed; eyes bright; lips and mucous membranes good color; pupils react to light. Chest well formed; percussion note clear in front and back; respirations clear in front and right back; a few sibilant râles heard over left back. Heart dulness somewhat increased to left. Apex beat to left of nipple line and forcible. Sounds are clear. Second pulmonic somewhat louder than second aortic. Pulse regular, 72. Abdomen is prominent in hypogastric and lower umbilical regions; wall held quite tense; right flank is slightly duller than left on

percussion and more painful on deep palpation; otherwise negative. Spleen not palpable. Urine, drawn by means of catheter, light yellow in color, alkaline in reaction, specific gravity 1011, contains much albumen, some red blood corpuscles, small number of pus cells, a little epithelium of transitional variety, but no casts. The specimen was centrifugalized.

Autopsy 12 hours after death. Anatomical diagnosis: Chronic diffuse nephritis (contracted kidney); slight heart hypertrophy; sero-fibrinous peritonitis; acute pleuritis; chronic pleuritis; pyosalpinx; swelling of follicles of intestine; single tuberculous ulcer of small intestine; chronic passive congestion and cloudy swelling of liver. Only such portions of the autopsy report will be given as bear on the present aspect of the case.

Peritoneal cavity.—In the dependent portion about 300 ccm. of faintly turbid serum containing flakes of fibrin. The peritoneum is congested, the vessels bright red and turgid, and small punctiform hemorrhages are visible beneath the peritoneal coat. Over the general visceral peritoneum there is a fine granular deposit of fibrin, which, on removal, leaves the underlying coat of the intestine free from gloss and vividly injected. Between the loops of the ileum, in the pelvis, are masses of fibrin easily removed by scraping with the finger. Over the stomach, spleen, liver, and on the under surface of the diaphragm is a similar thin fibrinous layer easily removable. The omentum is brightly injected. The body of the uterus is covered with a fibrinous membrane; on the ovaries, tubes and peritoneum covering the rectum are similar masses of fibrin, and in the latter larger hemorrhages exist.

Pleural cavity.—There is no excess of fluid in either. The lower lobe of left lung is covered by a fresh fibrinous deposit, and the same kind of membrane covers the pleural side of the diaphragm. No acute pneumonia.

Kidneys.—Combined weight, 155 grams. Dimensions, 9 × 5 × 2 cm. Average thickness of cortex, 4 mm. The capsule strips off with comparative ease, the surface is mottled and quite coarsely granular. On section the cortex is pale, the striæ obscure. The glomeruli are apparently diminished in number, but red. The cortex presents an opaque yellowish tint; the pyramids are darker in color than the cortex. Frozen sections show a great diminution in the number of uriniferous tubules and glomeruli in the specimens, and a corresponding increase in

the amount of intertubular tissue. The epithelium of the remaining convoluted tubules is quite fatty; and fat is also present in the glomeruli and newly-formed connective tissue. Many dense, hyaline casts are seen in the collecting tubules. There is an extensive obliterating endarteritis.

Intestine.—Just above the valve the Peyer's patches are swollen and congested, and an irregular ulceration 7.5 mm. in length exists at this place. The loss of substance does not extend beneath the submucosa. The solitary follicles of the small intestine are somewhat swollen. On microscopical examination the ulcer proves to be tuberculous. The base of the ulcer is covered with granulation tissue.

Bacteriological examination.—Cover-slip preparations made from the peritoneal exudate showed large bacilli only. From the organs negative. Cultures were made in agar-agar and gelatine from the peritoneal exudate in the pelvis, the peritoneum covering the diaphragm; the liver, spleen, kidneys and lungs. No growth was obtained from either the liver or spleen.

Peritoneal cavity.—The gelatine roll tubes from the pelvis contained after 24 hours in the thermostat a large number of small colonies; from the diaphragm similar but fewer colonies. Within the next 24 hours the former tube had run down, and in the latter the colonies had enlarged and now presented the characteristic swarming appearance of proteus colonies. Cover-slips from both tubes gave the same organisms: bacilli, with rounded ends, varying in size, and forms which resemble cocci. Slanting agar tubes were inoculated from a single colony from each of these sources. A grayish white carpet was obtained on the surface of the agar from each.

From these agar tubes dated November 3, 1892, gelatine Esmarch tubes were made on January 3, 1893. The agar tubes had dried down considerably, but the growth still seemed to be pure. These gelatine rolls were placed in the thermostat at 22° C. After 24 hours the series (three in number) from the pelvic peritoneal growth on agar, gave the following results: First tube very cloudy; there are large liquefying colonies on the surface and the tube is already beginning to run down. Under the low power of the microscope (Leitz No. 3 objective) many small colonies are visible in the depth of the medium causing the cloudy appearance

mentioned. The second tube contains fewer liquefying colonies; these have the size of a pin's head. Under the low power they are granular, quite circular in outline, and no projections are seen. Many small microscopic colonies also present. From the growth on agar from the diaphragmatic peritoneum a similar set of gelatine rolls was prepared. The growth here was a little slower and liquefaction less advanced than in the previous instance. Cover-slips from both sources show, however, the same bacteria to be present and these correspond with those previously described.

Hanging drop.—A single colony was examined and the organisms showed considerable motility. The rods looked a little longer and not quite so thick as in the specimen stained in carbol-fuchsin. The motion is either a swimming, side to side movement, or a rolling and tumbling one. Not all the bacteria were equally active, and some exhibited only the Brownian movement. The coccus-like forms presented a rapid, dancing movement. A large bacillus, 2 to 3 times the average length, with more sluggish movements, was observed. On January 5th the entire set of tubes had run down, and gelatine stab cultures from single colonies on these had, in 24 hours in the thermostat, grown out and a cup-like depression of liquefaction already made its appearance. Twenty-four hours later the liquefaction of the gelatine had progressed from above downwards so as to involve about half of the medium.

Milk.—January 8th two milk tubes, one plain and one with litmus, were made from the 24-hour old stab on gelatine from the peritoneum. Twenty-four hours later, the tubes having been kept at 37° C., the litmus milk was changed from pale blue to pink, but no coagulation had taken place. The plain milk was unchanged. After 48 hours the milk was still uncoagulated. The tubes were then removed from the thermostat and kept at the room temperature. They were not closely watched, and when looked at several weeks later both had coagulated, with an abundant separation of serum. The pink color of the litmus milk persisted.

Potato.—The surface growth after 48 hours was slight; but a more considerable growth had taken place in the fluid at the bottom of the tube. Cover-slips from the surface showed chiefly bacilli. They stained irregularly, and sometimes they were much distorted, evidently in process of involution.

Kidney.—A set of Esmarch roll tubes was prepared in gela-

tine from the original agar tube made at the autopsy. Placed in the thermostat at 22° C., tube number one was diffusely cloudy after 24 hours, and some large colonies were visible on the surface surrounded by a zone of liquefaction. Cover-slips showed bacillus and coccus forms, and a few of the bacilli were longer than the majority, equaling in length at times 4 to 6 of the average size. *In hanging drop* the motility resembled that described in the peritoneal tube. After 48 hours all the tubes had run down. A gelatine stab culture behaved in the same manner as those prepared from the peritoneal tubes. Transplantation to milk gave, after 48 hours, complete decolorization of the litmus, with the exception of a line corresponding with the separation of the cream where a pink color persisted. The milk was coagulated, with separation of serum. The plain milk tube was coagulated likewise.—*Potato.* After 24 hours there was a growth along the needle track, which after another day had increased. The color of the growth was grayish white. Cover-slips from this growth showed bacillus and coccus forms of micro-organisms, with the same variation in staining properties as has been mentioned in connection with the peritoneal potato growth.

Lung.—Gelatine Esmarch rolls show large colonies on the surface and smaller ones in the depth. The surface colonies were bluish in color, the margins serrated, and the whole colony seemed to project into the lumen of the tube. No tendency to liquefaction. Cover-slips from the two kinds of colonies show the same organisms: short, plump bacilli with rounded ends, 2 or 3 times as long as broad, with an occasional longer bacillus equal to 2 or 3 of the average size. Very seldom the bacilli were observed to be in pairs. Transplanted to *milk*, after 24 hours at 37° C. the plain milk apparently was unaltered; the litmus milk had assumed a pink color. After 48 hours both tubes were thickened, but no actual separation of serum had taken place. Removed from the thermostat and allowed to remain at the room temperature, after 8 hours the plain milk showed a beginning separation of serum, but the coagulum was finely subdivided. Less tendency to separation was manifested by the litmus milk. The tubes were not closely watched, but after several weeks at the room temperature they were again examined and the plain milk had coagulated firmly, the litmus milk somewhat less. On *potato* an abundant yellow growth was obtained.

We are, therefore, able to separate the organism found in the lung from those in the peritoneal cavity and kidneys. The first agrees with the bacillus coli communis, and as we have often found the colon bacillus in the lung in human beings dead of a variety of causes, we have no doubt as to its identity. The organisms from the peritoneal cavity and kidneys agree with each other in all essential particulars. It is worthy of note, however, that such a difference in the coagulating effect on milk exists, and this is of interest in connection with the known variation in the effect of the organism on gelatine. From the morphology and such of the biological properties of the organisms as we studied, it seems clear that they belong to the proteus group.

A word should, perhaps, be said with reference to the probability of the proteus having been a secondary invader, possibly post-mortem. We think that this supposition can be safely rejected. Although according to Bordoni-Uffreduzzi the proteus is the predominating post-mortem organism found in the tissues and blood of man and animals 24 to 48 hours after death, yet our experience has been far different. It is our rule to make cultures from all autopsies, and the proteus occurs rarely. Moreover, this autopsy was made in cold weather, soon after death, and no signs whatever of decomposition were present.

With reference to the pathogenic effects of proteus in this case there can be little doubt. An extensive peritonitis existed, and a single kind of organism was present in the exudate. Yet it must be considered that the natural properties of the proteus group of organisms are probably not such as to make them pathogenic for man under normal conditions. Hence it is of interest to consider just what the conditions may be in which they are enabled to exhibit such marked pathogenic properties as those mentioned. The proteus is a widely diffused organism in nature. It is found in the intestinal canal of human beings, and doubtless under ordinary conditions of health it might find its way into the peritoneal cavity without doing any great damage. But the conditions are greatly changed in such a case as this one. Here we have a system rendered particularly vulnerable to the invasion of bacteria in consequence of an extensive chronic disease. Cases somewhat similar are not rare. The terminal inflammations of serous membranes associated with long-standing renal and

hepatic disease are well known. But it is of especial interest to know that organisms which do not exhibit under ordinary circumstances any especial pathogenic properties in human beings, may, in the altered state of the fluids of the body caused by these diseases, produce definite and wide-spread pathological lesions.'

The present case furnishes an illustration in the existence of a chronic nephritis; and as an especial predisposing cause the ulcer of the intestine permitting a more ready ingress to the peritoneum from the intestine should be considered.

With reference to the classification of the several species of proteus originally described, Hauser now thinks that such a separation as first proposed is not justifiable. The distinction based on the liquefaction of gelatine has been found to be insufficient to distinguish the several kinds, for in the case of a specimen of P. Zenkeri which had during 60 generations not liquefied gelatine, the 61st generation caused its liquefaction. Hence he now regards the several species originally made as varieties of proteus vulgaris.

Kuhn had previously come to the conclusion that P. mirabilis could not be considered a distinct species, but still adhered to the difference between P. vulgaris and P. Zenkeri.

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